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US ARMY MEDICAL RESEARCH LABORATORY

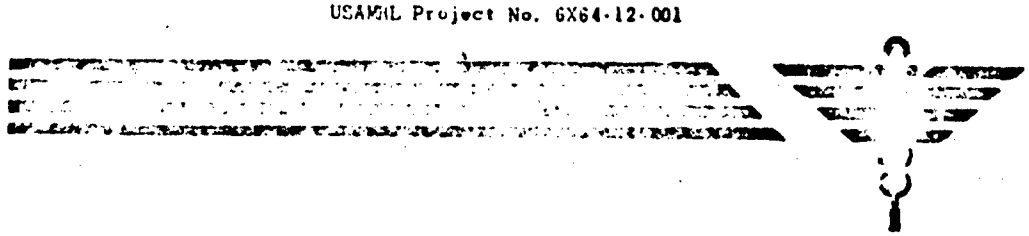
FORT KNOX, KENTUCKY

REPORT NO. 509

HEMODYNAMICS OF THE STOMACH
III. EFFECTS OF SALMONELLA TYPHOSA ENDOTOXIN ON THE RESISTANCE TO BLOOD FLOW

E. S. Dooley, Ph. D.
J. B. Scott, M. S.
Capt E. D. Frohlich, MC
Capt E. D. Jacobson, MC

Studies of Physiological Effects of Cold on Man
Task 01
Environmental Medicine
USAMRL Project No. 6X64-12-001



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Task 01
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Report No. 509
USAMRL Project No. 6X64-12-001-01

ABSTRACT

HEMODYNAMICS OF THE STOMACH
III. EFFECTS OF SALMONELLA TYPHOSA ENDOTOXIN ON THE RESISTANCE TO BLOOD FLOW

OBJECT

This study was designed to determine the response of the gastric vascular bed to the local administration of the endotoxin of Salmonella typhosa 0901.

RESULTS

The injection of endotoxin into the left gastric artery of 10 dogs produced a rapid average increase in gastric arterial pressure (100%) and coronary venous pressure (200%). Arterial pressure remained elevated for 30 minutes, but venous pressure returned to control in 15 minutes. Systemic arterial pressure fell an average of 20% in 10 minutes. Locally infused phentolamine blocked the responses of the gastric artery and coronary vein without affecting systemic pressure.

In a second series of animals whose systemic and gastric circulations were completely separated, endotoxin administered into either the gastric or systemic circulation failed to produce rapid increases in gastric vascular resistance.

CONCLUSIONS

These studies indicate that the left gastric arterial and coronary venous pressure increases induced by endotoxin in the intact animal are probably mediated in large part by systemic release of catecholamines.

RECOMMENDATIONS

None.

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HEMODYNAMICS OF THE STOMACH
III. EFFECTS OF SALMONELLA TYPHOSA ENDOTOXIN ON THE RESISTANCE TO BLOOD FLOW

I. INTRODUCTION

Endotoxin is responsible for many profound vascular effects including venous pooling, changes in resistance to blood flow in many organs, and circulatory collapse leading to death. Most prominent among the many early vascular effects of endotoxin is the rapid development of hepatic and intestinal congestion (1, 2). The apparent cause of this pooling of blood in these organs is venous constriction in the liver. With time hepatic engorgement subsides, but pooling of blood in the intestine continues, suggesting some persistent effects of endotoxin on the blood vessels of the gut (2).

The gastric circulation, like the intestinal, is in direct continuity with the portal vein and might be expected to exhibit responses similar to those of the gut vasculature. This investigation is concerned with the response of the gastric vascular bed to the local administration of the endotoxin of Salmonella typhosa 0901.

II. METHODS

Twenty-five mongrel dogs of both sexes weighing eight to 20 Kg. were subjects of acute studies. The animals were anesthetized with pentobarbital sodium (35 mg. per Kg.) and anticoagulated with heparin sodium (5 mg. per Kg.). Artificial respiration was administered by a tracheal cannula when required. The stomach and its main blood vessels were exposed by a left subcostal incision. Splenectomy was routinely performed and the stomach was ligated at both ends. Under these conditions the vagi were probably not functioning.

Following ligation of the hepatic and splenic arteries near their origins, the right gastric and right gastroepiploic arteries and veins, and the multiple branches of the left gastroepiploic artery and vein which enter and leave the greater curvature were ligated. Blood was perfused through the left gastric artery of the stomach by a pressure-independent, variable flow pump (Sigmamotor Pump, Model T-65) interposed between the right femoral artery and the celiac axis.

Needles were inserted into the coronary vein of the stomach, the perfusion tubing proximal to the left gastric artery, and the left common

carotid artery and connected to a strain guage for recording of blood pressures (Sanborn Twin-Viso, Model 60).

Salmonella typhosa 0901 endotoxin (0.6 mg. per Kg. Bacto Lipopolysaccharide, Difco Laboratories) was injected in one bolus directly into the perfusion system just proximal to the left gastric artery.

Blood flow through the gastric artery was fixed at a given rate for each experiment. Flow varied from 24 to 48 ml. per minute.

Single Circulation. Pressures were recorded from five minutes before to 30 minutes after injecting endotoxin in ten dogs. In three additional animals this experiment was repeated, but a phentolamine infusion in doses which did not alter systemic arterial pressure (0.5 to 3.0 µg. per minute) was administered into the gastric artery throughout the experiments. In three other animals no endotoxin was administered and the results obtained for 30 minutes in these dogs served as control records. In these latter three control animals the systemic arterial pressure was lowered by exsanguination to the same degree as observed with endotoxin and the effect on the gastric arterial pressure was measured.

Dual Circulations. In nine other animals venous drainage from the stomach was collected in a reservoir connected to a pump-oxygenator (Kay-Cross disc oxygenator, Pemco Inc.) before being returned to the left gastric artery. This constituted a closed circuit gastric circulation. Endotoxin was injected into the perfusion system as outlined above in three of these nine dogs and injected into a systemic vein in another three animals of this group. No endotoxin was given to the remaining three dogs which served as controls. Recordings were obtained in these nine dogs for the subsequent 30 minutes. The isolation of the gastric circuit was checked by two methods: the volume of blood collected from the coronary vein was compared with the volume perfused through the gastric artery, and India ink was injected into the perfusate to determine whether leakage of ink was occurring beyond the confines of the stomach. These procedures showed that the stomach circulation was isolated from the systemic.

III. RESULTS

Single Circulation. The injection of endotoxin into the gastric artery of ten dogs caused a rise in coronary venous pressures followed within two minutes by an increase in left gastric artery pressures and a decrease in left common carotid artery pressures. Peak venous

pressures were achieved at about five minutes after injection and averaged over 200 per cent of control. Venous pressures then returned to pre-injection levels over the next 15 minutes. Left gastric artery pressures continued to climb to maximum values for ten minutes beyond injection and then decreased, but were still nearly twice control values at 30 minutes. The pressure gradient across the gastric vascular bed showed a mean increase at ten minutes of 107 per cent and at 30 minutes was 89 per cent above the pre-injection value. These values were significant ($p = <.001$) when compared with pressure gradient changes obtained in the three control animals using the Student t - test (3). Systemic pressures reached a nadir at ten minutes after injection of endotoxin when an average value of 24 per cent below the pre-injection level was recorded. Subsequently, pressure in the carotid artery returned to a mean value of 13 per cent below initial pressure at 30 minutes after injection. These results appear in Fig. 1.

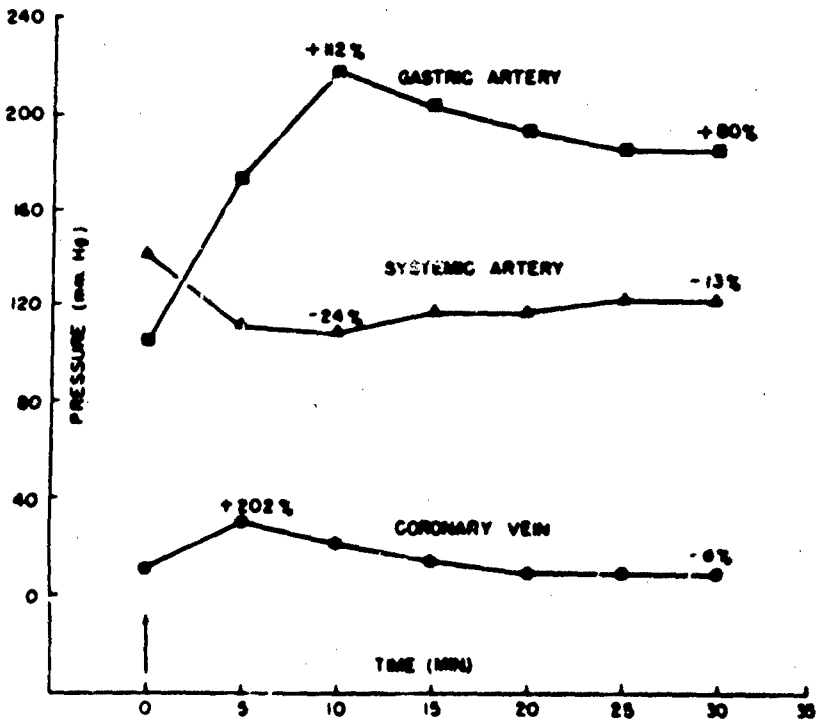


Fig. 1. Average pressures from ten dogs in whom endotoxin was injected into the left gastric artery (arrow). Gastric and systemic circulations were in continuity. Per cent change from initial values is noted at various times.

In the three animals in whose perfusion system a phentolamine infusion was maintained after injection of endotoxin, the gastric arterial pressure increased only 18 per cent over the pre-injection value. Venous pressure rose only 30 per cent in these dogs. The results from these animals are shown in Table I.

In the three control animals the mean gastric artery pressure varied by no more than 13 per cent above or below initial pressure over a period of 30 minutes. The coronary vein and systemic arterial pressures displayed correspondingly small fluctuations. These results are shown in Table I.

Lowering systemic blood pressure 20 per cent by bleeding had no effect on gastric artery pressure in the three control animals.

Dual Circulations. In the nine animals in whom the circulation of the stomach was separated from the systemic circulation and maintained with a pump-oxygenator, the venous pressure was atmospheric throughout. In the three dogs of this group in whom no endotoxin was administered, there was a progressive rise in mean left gastric artery pressure of 63 per cent over 30 minutes, while systemic arterial pressure varied by no more than 15 per cent above or below control. The results from these three control animals used to show the effect of time on gastric artery pressure are seen in Table II.

In the three dogs of this group in whom endotoxin was injected into the gastric circulation and did not reach the systemic circulation, there was no increase in perfusion pressure. At 30 minutes gastric arterial pressure was the same as at the time of injection and was significantly lower than that found in the three control dogs with separate circulations ($p = <.02$). Systemic pressure fluctuations were the same as in the control animals. These results are shown in Table II.

In the three dogs with dual circulations in whom endotoxin was injected into the systemic circulation, pressures at 30 minutes were not significantly different from the dual circulation control animals. These results are shown in Table II.

The effect of endotoxin on gastric vascular resistance was calculated for the six series of experiments: control, endotoxin, and endotoxin with phentolamine in the animals with a single circulation, and control, endotoxin in the gastric artery and endotoxin in the systemic vein in the dogs with two separate circulations. It can be seen (Fig. 2a)

TABLE I. PRESSURE CHANGES IN THE LEFT GASTRIC ARTERY, THE CORONARY VEIN AND LEFT COMMON CAROTID ARTERY IN 3 CONTROL ANIMALS AND 3 ANIMALS GIVEN ENDOTOXIN WHILE PHENTOLAMINE WAS INFUSED. THE GASTRIC CIRCULATION WAS IN CONTINUITY WITH THE SYSTEMIC IN BOTH SERIES.

			CONTROL																			
			Pressure (mm. Hg)																			
Dog No.	Flow (ml./min.)	Min.	0	5	10	15	20	25	30	0	5	10	15	20	25	30	0	5	10	15	20	25	30
11	41		125	148	140	140	146	152	156	15	20	22	22	23	23	19	144	160	160	160	160	156	154
12	34		140	130	120	124	128	122	124	15	17	17	18	17	16	16	132	136	144	144	144	132	136
13	34		140	140	126	110	120	110	128	17	16	16	16	20	22	20	144	148	138	144	144	144	140
Mean	36		135	139	129	125	131	129	136	16	18	18	19	20	20	18	140	148	147	149	149	144	143
			PHENTOLAMINE AND ENDOTOXIN																			
14	35		128	142	140	124	108	116	120	12	18	18	9	7	5	5	126	116	104	100	98	92	94
15	28		86	96	108	132	140	150		8	12	10	10	10	10		124	126	120	120	112	112	
16	41		160	144	176	184	152			19	23	23	15	17			136	116	138	130	108		
Mean	35		125	127	141	147	133	135	120	16	21	17	11	11	8	5	129	119	121	117	106	102	94

TABLE II. PRESSURE CHANGES IN THE LEFT GASTRIC ARTERY IN 3 SERIES OF EXPERIMENTS IN WHICH THE GASTRIC CIRCULATION WAS ISOLATED FROM AND INDEPENDENT OF SYSTEMIC CIRCULATION. CORONARY VENOUS PRESSURE WAS ATMOSPHERIC.

			CONTROL						
Dog No.	Flow (ml./min.)	Min.	Left Gastric Artery Pressure (mm. Hg) 0	5	10	15	20	25	30
17	41		248	280	320	344	364	384	424
18	41		226	308	304	312	340	348	352
19	42		208	220	260	288	304	328	336
Mean	41		227	269	295	315	336	353	371
			ENDOTOXIN IN THE GASTRIC CIRCULATION						
20	27		160	152	164	180	176	174	180
21	31		264	264	264	272	272	272	272
22	42		336	304	308	312	304	308	304
Mean	33		253	240	245	255	251	251	252
			ENDOTOXIN IN THE SYSTEMIC CIRCULATION						
23	25		180	216	240	260	262	272	276
24	28		104	128	124	136	148	164	180
25	27		256	264	264	272	280	280	264
Mean	26		180	203	209	222	230	238	248

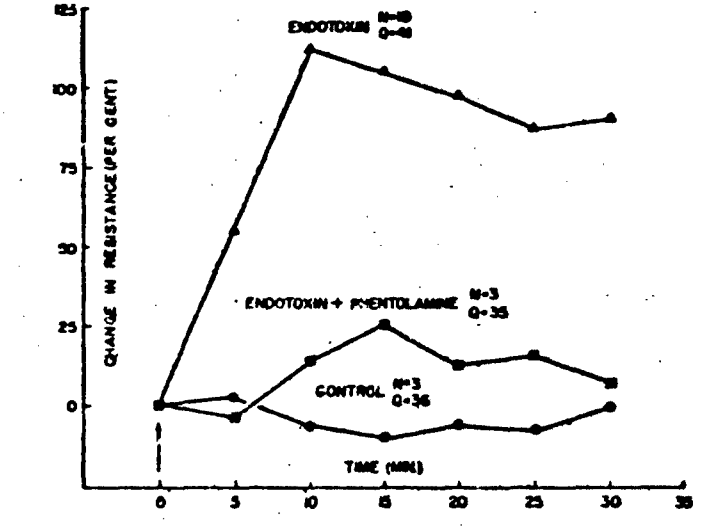
that endotoxin induces a persistent increase in gastric vascular resistan which is blocked by phentolamine infused into the gastric artery. In the animals with separate circulations (Fig. 2b) endotoxin injected into the gastric circulation prevented the rising resistance observed in the contr dogs, while endotoxin in the systemic circulation induced no significant change from control.

IV. DISCUSSION

Endotoxin when injected directly into the left gastric artery and allowed to circulate systemically produced a prolonged increase in gast vascular resistance. This effect was abolished by phentolamine. When the gastric circulation was isolated from the systemic and endotoxin injected into the systemic circulation there was no change from control. When endotoxin was injected into the separate gastric circulation the resistance increase, which had been noted in the three control animals, d not occur. These studies suggest that endotoxin produces an active increase in the gastric vascular resistance in the dog by an indirect mech nism. This increase is produced, at least in part, by a remote release of catecholamines into the blood. It also appears that the direct effect of endotoxin on the isolated gastric circulation is a prevention of the increase in gastric vascular resistance observed in the control dogs with dual circulations. In the animal with a single circulation, however, the indirect mechanism predominates.

Several factors might have been responsible for the increase resistance to blood flow in the stomach of the single circulation animals i ducted by endotoxin. The rise in venous pressure, presumably due to hepatic venous congestion, might have actively elevated resistance in th stomach. Endotoxin when administered into the left gastric artery migl have acted directly on the gastric vasculature or have liberated a vasoactive chemical mediator locally; or it might have acted indirectly upon a distant site by nervous or chemical means.

It appears unlikely from these studies that the elevation of venous pressure in the stomach could have had more than a minor role in raisi gastric arterial pressure. Peak coronary vein pressures were reache long before arterial pressures were maximal, and pressures in the left gastric artery remained twice control long after restoration of venous pressures to control values. The veno-arteriolar reflex, as described in other beds (4, 5), exerts a nearly immediate effect rather than the sequence observed here. In preliminary investigations in this laborato a three-fold rise in coronary venous pressure was not accompanied by



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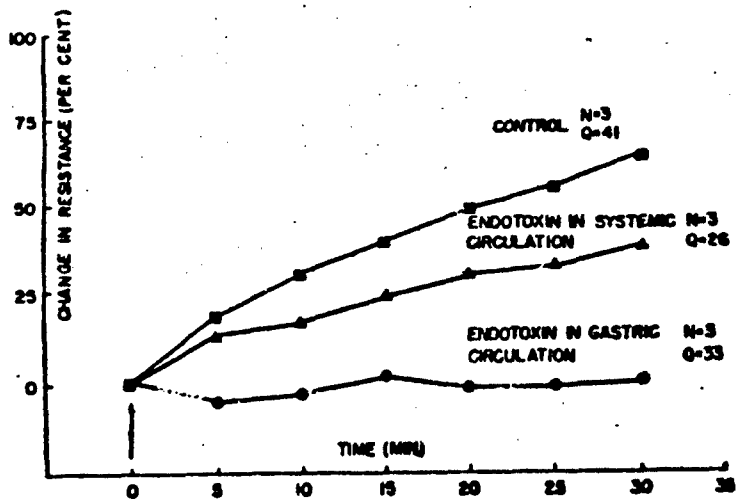

b

Fig. 2. Changes in gastric vascular resistance in the six series of experiments. Figure a compares resistance changes in the single circulation experiments in the control dogs with changes induced by endotoxin and by endotoxin and phentolamine. Figure b compares resistance changes in the dual circulation experiments in the control series with the groups given endotoxin either in the gastric artery or systemic vein. N signifies the number of subjects, Q represents the mean flow for the series (ml. per min.) and the arrow indicates endotoxin injection.

increases in gastric arterial pressures which were of the magnitude observed after injecting endotoxin.

When endotoxin was injected into the isolated gastric circulation, resistance remained unchanged for the subsequent 30 minutes. In the control animals with two circulations resistance increased 63 per cent in 30 minutes (Fig. 2b). This suggests that the local action of endotoxin in the gastric vascular bed is dilatation.

The increase in gastric vascular resistance must be secondary to a distant mechanism of endotoxin. This inference is confirmed by the three experiments with locally infused phentolamine and the studies with separate circulations in which endotoxin was excluded from the stomach circuit. Phentolamine was infused into the left gastric artery in amounts which did not affect systemic arterial pressure. The failure of endotoxin to elevate pressure significantly in the gastric artery, where phentolamine concentration was high, suggests that circulating catecholamines or the sympathetic nervous system was responsible for the increases in gastric artery pressure due to endotoxin.

In the three double circulation experiments in which endotoxin was injected systemically and presumably neither endotoxin nor any substances it elaborated could have reached the gastric circulation, gastric arterial pressures were not significantly different from the control animals with two circulations (Fig. 2b). This suggests that endotoxin acts to raise gastric vascular resistance in the intact animal through local vasoconstriction mediated primarily by circulating substances. The sympathetic nervous system is of little importance locally in this preparation.

The mild early systemic hypotension induced by endotoxin was not responsible for the gastric vascular resistance changes. When comparable degrees of hypotension were induced by exsanguination, left gastric artery pressure did not change.

The nature of the mechanism whereby endotoxin elaborates vasoactive substances is not evident from these studies.

V. SUMMARY

The effects of endotoxin on the gastric vascular bed of the dog were investigated using an acute preparation in which flow was kept constant and pressure allowed to fluctuate freely. Both gastric artery and coronary venous pressures exhibited marked rapid increases in response to S. typhosa endotoxin administered into the left gastric artery in ten dogs.

Arterial pressure doubled and venous pressure increased by about 200 mm. Hg within five to ten minutes after endotoxin injection. Arterial pressure remained elevated for 30 minutes, but venous pressure returned to normal in 15 minutes. Simultaneously, systemic pressure fel 20 per cent in ten minutes and only partly recovered by 30 minutes. Th responses of the gastric artery and coronary vein were blocked by local infused phentolamine.

In a second series of animals whose systemic and gastric circulations were completely separated, endotoxin administered into the gastric circulation prevented the resistance increase observed in control animals and in animals with endotoxin administered in the systemic circulation. The response of the gastric vasculature to endotoxin noted in the intact animals was not observed in the dogs with separate gastric and systemic circulations in whom endotoxin had been administered in either circulation.

These studies indicate that the left gastric arterial and coronary venous pressure increases induced by endotoxin in the intact animal are mediated by increased levels of circulating catecholamines.

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